EPA Comments on Chemical RTK Challenge Submission:

2,4-Di-*tert*-butylphenyl phosphite (3:1)

SUMMARY OF EPA COMMENTS

The sponsor, Ciba Specialty Chemicals Corp., submitted to EPA Robust Summaries and a Test Plan that were received July 10, 2000, for 2,4-Di-*tert*-butylphenyl phosphite (3:1) (CAS # 31570-04-4). EPA posted the submission on the ChemRTK website on July 20, 2000.

EPA has reviewed this submission and has reached the following conclusions:

1. The submission does not meet minimal test plan standards for data adequacy. There were many inadequacies in the study summaries, which need revision to be acceptable for the Challenge Program. EPA has provided specific comments on how to enhance the robust summaries. Sponsors should refer to the Challenge Program guidance.

EPA accepts the submission conditionally, believing that the issue is poor documentation but that enough information may be inferred to make tentative judgements. Eventual full acceptance of the submission is contingent upon the receipt within 90 days of substantially improved robust summaries and other information that can meet the standard set out in EPA's guidance documents.

2. Physicochemical properties and environmental fate. The calculated data in the submission are generally satisfactory. However, EPA recommends that, when possible, measured data should be used; in this case, a measured melting point has been reported. To estimate transport and distribution, the sponsor has used the EPIWIN Level III model which provides estimated values as inputs; here again, EPA recommends using measured data whenever possible. EPA further recommends using the EQC level III model from the Canadian Environmental Modeling Centre at Trent University.

3. Health Effects. Most of the robust summaries are inadequate because not enough information is presented to allow for an independent assessment of the data. However, EPA's tentative scientific judgment is that no further testing is needed for the purposes of the U.S. HPV Challenge Program, provided that the sponsor supplies adequate documentation as discussed under Item 1 above.

4. Ecological effects. Although there were many inadequacies in the study summaries, EPA suggests that an analysis based on this chemical's physicochemical properties, including extremely low water solubility, may support the sponsor's conclusion that no further testing is necessary. EPA will take into account adequate documentation of such an analysis supplied by the sponsor in determining final acceptance of the test plan.

EPA is requesting that the Sponsor advise the Agency within 60 days of any modifications to its submission.

EPA COMMENTS ON THE 2,4-DI-*tert*-BUTYLPHENYL PHOSPHITE (3:1) CHALLENGE SUBMISSION

General

The submission does not meet minimal standards. There were many inadequacies in the health and ecological effects study summaries, which must be revised to allow final acceptance as an HPV Challenge submission. EPA has provided specific comments on how to enhance the robust summaries to the standard established in EPA's HPV Challenge Program Guidance (http://www.epa.gov/opptintr/chemrtk/guidocs.htm).

Test Plan

Chemistry (melting point, boiling point, vapor pressure, water solubility, and partition coefficient).

EPA believes that no additional test data are needed to satisfy the needs of the HPV Challenge Program.

Fate (photodegradation, stability in water, biodegradation, and transport/distribution).

EPA believes that no additional test data are needed to satisfy the needs of the HPV Challenge Program.

Health Effects (acute toxicity, repeat dose toxicity, genetic toxicity, and reproductive/developmental toxicity).

EPA's tentative judgement is that no additional test data are needed to satisfy the needs of the HPV Challenge Program pending receipt of adequate robust summaries.

Ecological Effects.

EPA's tentative judgement is that no additional test data are necessary to satisfy the needs of the HPV Challenge Program pending receipt of adequate robust summaries. An adequately documented analysis, such as a quantitative structure-activity relationship (QSAR) analysis, based on this chemical's physicochemical properties may provide additional support for the sponsor's conclusion that further aquatic testing is unnecessary.

SPECIFIC COMMENTS ON ROBUST SUMMARIES

Chemistry

All the physicochemical (P/C) data reported in the Robust Summary are estimates from the EPIWIN program. Since this chemical has extremely low water solubility and vapor pressure, it is reasonable to present calculated values for these endpoints. EPA found no existing P/C data for this chemical except a published melting point of 181–184 °C (Aldrich Catalog) (for chemicals with a molecular weight greater than about 200 or with more than 15 carbons, EPIWIN almost always predicts a melting point much higher than observed). Recalculation in EPIWIN using this melting point yielded essentially no change in boiling point and Log Kow. Vapor pressure (5.3×10^{-13} torr @ 25 °C) and water solubility (4.8×10^{-15} mg/L @ 25 °C) were about 10-fold higher and lower, respectively. In practical terms, the vapor pressure and water solubility values are so low for this chemical by either calculation that they can be considered negligible.

Nonetheless, as measured values are preferred as inputs to other estimation programs, sponsors should explain their use of an estimated value when a measured value is available.

Fate

The environmental fate data summaries have been reviewed and are satisfactory. However, measured input values are preferred for the transport/distribution modeling if available, and the estimated melting point value of 268 C should be replaced by the published value of 181–184 °C, although in practical terms this substitution does not change the modeling in a significant way in this case.

EPA recommends using measured data as much as possible. The sponsor used the EPIWIN Level III model, which provides estimated values as default inputs. In order to estimate environmental fate endpoints, however, EPA recommends using the EQC level III model from the Canadian Environmental Modeling Centre at Trent University. This model can be found at the following website: http://www.trentu.ca/academic/aminss/envmodel/.

Health Effects

EPA evaluated thirteen health endpoint robust summaries and determined that ten were inadequate summaries for the purposes of the U.S. HPV Challenge Program. The inadequate summaries are lacking information that is necessary to evaluate the basic adequacy of the cited study. EPA's tentative scientific judgment is that new studies are not necessary for the purposes of the U.S. HPV Challenge Program, pending receipt of adequate robust summaries.

The following EPA comments reflect the information in the robust summaries (the full study reports may address these comments):

Acute Toxicity: The acute oral toxicity study in rats: a 7 day post-exposure observation period was used instead of 14 days. In both the acute oral and dermal studies: because a vehicle was used, a control

should have been run (but since no deaths occurred, a vehicle effect was not likely).

Repeat Dose Toxicity. Three separate robust summaries were submitted for this endpoint. Comments on the 90-day rat study: (1) it is important to report the incidence of the increased kidney weights in the females during the study; and (2) it is important to know whether this effect was reversible in the 28-day post-exposure satellite group. Comment on the 90-day dog study: the sponsor states that there was no significant effect on body weight or clinical chemistries (including hematology and urinalysis). It is important to know the incidence of any effects observed because it may be biologically relevant even though it is not statistically significant.

Genotoxicity (somatic mutation assay [SCE study in bone marrow cells], Chinese hamsters). (1) Did not report the use of a positive control. (2) While a positive response was observed at the highest dose, the incidence (number of SCEs per cell of treated vs. control) was not reported. (3) In addition, it is not clear why the study was repeated in 1989 at higher dose levels than the original study performed in 1982.

Genotoxicity (chromosome study in spermatocytes of mice). The in vivo study with male mice (assessing chromosome aberrations in primary and secondary spermatocytes) is considered inadequate because, according to OECD 483 (chromosome aberration test guideline for spermatogonia), animals should be sacrificed 24 hours after one or two doses in order to capture the meiotic cycle properly. The robust summary reported that five doses were given and sacrificing occurred three days after the last dose. In addition, there are two references for this study (1982 and 1989) - was the test repeated?

Genotoxicity (somatic mutation assay [nuclei anomalies in bone marrow cells], Chinese hamsters). (1) No data are presented to support the claim that there is no difference between treated and control cells in numbers of anomalies.

Genotoxicity (dominant lethal assay, mice). (1) No positive or negative controls were reported. (2) There is no indication of male:female mating ratio. (3) In addition, although the statement is made that no evidence of dominant lethality was observed, no supporting data are presented.

Genotoxicity (Ames test). (1) There is no rationale given for dose selection; (2) the background revertant colony counts are not reported; (3) there is no indication whether positive and negative controls responded appropriately; (4) there is no indication whether a solvent or vehicle was used; (5) there is no indication of incubation temperature or method of counting, e.g, by hand or electronic colony counter; and (6) no data are presented to support the claim that there is no increase in reverse mutation with or without S9 fraction.

Genotoxicity (yeast cells). The in vitro test with yeast cells is considered inadequate for the following reasons: (1) no mammalian metabolic activation system was used; (2) the yeast strain used (MP-1) is not on the OECD 480 test guideline list of appropriate strains; (3) OECD 480 suggests incubations of up to 18 hours at 28-37 ^{o}C, instead of the 3.5 hours at 25 ^{o}C reported in the robust summary; and (4) no data are presented supporting the statement that the test material was non-mutagenic.

Reproductive Toxicity Because no details were provided (likely due to no effects being observed except for a reduction in body weight of F0 females in the two-generation study at the highest tested dose of 10,000 ppm of the test material in the diet, or approximately 500 mg/kg), it is not possible to make an independent assessment of the test results. It is necessary to provide the actual data (incidence by dose) for the parameters assessed. This could be in the form of a summary table.

Developmental Toxicity Because no details were provided, it is not possible to make an independent assessment of the test results. It is necessary to provide the actual data (incidence by dose) for the parameters assessed. This could be in the form of a summary table.

Ecotoxicity Studies

The comments below reflect the information presented in the robust summaries; information in the full study report may address some of the issues identified.

Acute Aquatic Toxicity. Robust summaries were submitted for studies on fish, daphnia, and green algae. Many critical experimental details were omitted from the robust summaries which effectively rendered them inadequate. EPA's tentative scientific judgment is that new studies are not necessary for the purposes of the U.S. HPV Challenge Program, pending receipt of adequate robust summaries and adequate analysis of potential effects.

Fish. Information missing from the robust summary includes: test substance purity, pH, TOC, dissolved oxygen, hardness, and concentration of acetone vehicle. Test concentrations were above the predicted

water solubility limit; and there was no information as to whether the test concentrations were measured or nominal. Because this chemical has low water solubility, the information provided does not allow a conclusion as to how much of the test was conducted according to the Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures (OECD, June 2000 available at http://www.oecd.org/ehs/test/monos.htm). Other submitted fish LC50 data did not have robust summaries to support the observed moderate toxicities.

Aquatic plants. Information missing from the submitted algal inhibition test robust summary includes: test substance purity, total hardness, pH, TOC, exposure vessel type, size, lighting, temperature, vehicle concentration and dissolved oxygen. The chemical was tested above the calculated water solubility limit. The test was conducted using unknown concentrations of Tween 80 to solubilize the chemical. This solvent is not generally advocated because it may exert physical toxicity and interfere with test substance concentration measurement. For more information refer to the OECD Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixtures.

Aquatic invertebrates. Information missing from the submitted acute daphnid test robust summary includes: test substance purity; a description of the dilution water to include source, temperature, dissolved oxygen, pH, hardness, alkalinity, and total organic carbon; vehicle; and vehicle concentration. The summary did not specify whether concentrations were measured or nominal. The chemical was tested above the calculated water solubility limit, and test duration was only 24 hours instead of the recommended 48 hours.

Followup Activity

EPA requests that the Sponsor submit adequate robust summaries and other modifications to its submission within 90 days.